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# Developmental Biology

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## Concurrent Session 8: Branching and Migration

### Program/Abstract # 54

#### The role of the apical PAR-polarity complex in branching morphogenesis

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How cells form their complex three-dimensional morphologies is a major unsolved problem in cell and developmental biology. To address this, we are studying the development of a specific cell type, the *Drosophila* tracheal terminal cell, a component of the insect respiratory system. Each terminal cell forms dozens of fine subcellular branches by a process of cytoplasmic extension and bifurcation, followed by tubulogenesis, in which a subcellular lumen is formed within all the branches. To identify the molecules required for development of terminal cells, we performed a large scale genetic screen. One of the mutants we obtained shows highly reduced cell branching, as well as defects in lumen formation. We have found that this mutation is in the cell polarity gene *par-6*, a member of the apical PAR-polarity complex. This complex is best known for maintaining apical/basal polarity within epithelial cells and for being required for many asymmetric cell divisions. We have found that another member of the apical-polarity complex, *bazooka* (the *Drosophila* homolog of *par-3*), is also required for terminal cell branching, but surprisingly is not required for lumen formation. Our results suggest two hitherto undescribed aspects of the apical-polarity complex. First, the complex plays a role in specifying another kind of regional difference within cells: the determination of branch points in a branching cell. Second, in most epithelia, cell junctions are required along with the apical-polarity complex to maintain polarization. Terminal branches contain no such junctions, suggesting a novel mechanism of establishment or maintenance of apical-basal polarity.

doi:[10.1016/j.ydbio.2010.05.088](https://doi.org/10.1016/j.ydbio.2010.05.088)

### Program/Abstract # 55

#### FGF-induced collective cell migration during lung branching morphogenesis

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Vertebrate lung forms tree-like structure by repeated branching of epithelial tip during development. We previously formulated a mathematical model of lung branching morphogenesis based on FGF-induced growth, but actually it has been shown that FGF10, which is produced by the mesenchyme surrounding epithelial tip, acts as a chemoattractant in this process. In *Drosophila* airway system, leading cells become mesenchyme-like and extend filopodia to migrate, but mouse lung epithelium moves toward FGF source

while retaining epithelial integrity. Cell migration in single cell level has been extensively studied, but how the sheet-like epithelial structure moves toward chemoattractant source remains to be elucidated. In the present study, we developed a primary cell culture system of developing lung epithelium and combined it with baculovirus-based transfection system to visualize the cytoskeleton dynamics. We found that tightly packed epithelial cells can protrude lamellipodia or filopodia underneath the neighboring epithelial cells. The observed structure has been described as “cryptic lamellipodia” in MDCK cell line. As a result, cytoskeleton of each epithelial cell shows dynamics very similar to that of single cell movement. Distinction between chemotaxis-induced and cell proliferation-induced branching morphogenesis in comparison with mathematical models is discussed.

doi:[10.1016/j.ydbio.2010.05.089](https://doi.org/10.1016/j.ydbio.2010.05.089)

### Program/Abstract # 56

#### Dynamic in vivo multispectral imaging and cell tracking of chick neural crest cell migration using multiple histone fluorescent proteins

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Neural crest migration is a highly dynamic event in the vertebrate embryo that involves cell-to-cell contact and long distance travel. How neural crest cells (NCCs) communicate positional information and coordinate collective movements to ensure discrete migratory streams reach precise targets is not well understood. The detailed examination of this question involves studying cell-to-cell contact dynamics and accurate cell tracking. In the past, we have lacked the tools to resolve overlapping cellular processes and trajectories in dense populations of migratory cells in vivo. Here, we report that using multiple nuclear localizing histone fluorescent proteins (H2B-GFP, H2B-YFP and H2B-mCherry) leads to more accurate tracking of single NCC trajectories by spectral identity through wavelength fingerprinting. We also show that by using combinations of multiple membrane and nuclear localizing fluorescent proteins (Gap43-GFP, Gap43-YFP and H2B-mCherry), we are able to detect and separate individual NCC filopodial processes in vivo. We present measurements of the number, duration, and surface area of cell-to-cell contacts in a typical NCC migratory stream and highlight differences based on cell position within the stream. In summary, our multicolor, multispectral in vivo imaging approach reveals new details of neural crest migration in unprecedented detail.

doi:[10.1016/j.ydbio.2010.05.090](https://doi.org/10.1016/j.ydbio.2010.05.090)